

Blood Washing Technique for Fresh Blood

I. Getting Red Blood Cells from donor

- 1) Divide the blood into 50 ml fractions in 50 ml Falcon tubes.
- 2) Place blood at 4 °C for **AT LEAST** 48 hours.
- 3) Collect the upper serum layer (approx. half the tube) and save the packed red blood cells at 4 °C. (**The RBCs are good for ONLY ONE MONTH!**)
(Save the serum in a 250 ml conical tube and treat using the Serum Collection Protocol)

II. Red Blood Preparation for Malaria culture

- 1) Put 5-10 ml of packed RBCs into 50 ml Falcon tubes and fill to 45 ml with RPMI 1640 media (ours contains 2.0 g/L glucose, 2.0 g/L NaHCO₃, 0.3 g/L L-Glutamine)
- 2) Mix well by gently shaking.
- 3) Spin down in table-top centrifuge at 1500 RPM for 10 min at 20 °C (NO BRAKE).
- 4) Aspirate off the supernatant.
- 5) Mix an equal volume of RPMI to the RBCs to make a 50% mixture.
- 6) Store in the fridge at 4 °C not more than 4 days. (It is always better to make it fresh!)